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SEQUENCE LISTING



<110> Bruck, Claudine
Bollen, Alex
Jacobs, Paul
Massaer, Marc

<120> Recombinant Allergen with Reduced
Enzymatic Activity

<130> B45122

<140> 09/554,860

<141> 2000-05-19

<150> PCT/EP98/07521

<151> 1998-11-16

<150> GB 9724531.0

<151> 1997-11-19

<160> 30

<170> FastSEQ for Windows Version 4.0

<210> 1

<211> 963

<212> DNA

<213> Artificial Sequence

<220>

<223> Mutant of DerP1

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gaagatgaag aagctgcccg taaaaacttt ttggaatcag taaaatatgt tcaatcaaat 180
ggaggtgccca tcaaccattt gtccgatttg tcgttgatg aattcaaaaa ccgatttttg 240
atgagtgcag aagcttttga acacctcaaa actcaattcg atttgaatgc tgaaactaac 300
gcctgcagta tcaatggaaa tgctccagct gaaatcgatt tgcgacaaat gcaactgtc 360
actccattc gtagtcaagg aggctgtggt tcagcttggg ctttctctgg tgttgccgca 420
actgaatcag cttatttggc ttaccgtaat caatcattgg atcttgctga acaagaatta 480

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tgccgacgac caaatgcaca acgtttcggg atctcaaact attgccaaat ttaccacacca 660
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ggcatcaaag atttagacgc attccgtcat tatgatggcc gaacaatcat tcaacgcgat 780
aatggttacc aaccaaacta tcacgtctgc aacattgttg gttacagtaa cgcacaaggt 840
gtcgattatt ggatcgtacg aaacagttgg gataccaatt ggggtgataa tgggttacggt 900
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963

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<210> 2

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<213> Artificial Sequence

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<400> 2

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Met Lys Ile Val Leu Ala Ile Ala Ser Leu Leu Ala Leu Ser Ala Val
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Tyr Ala Arg Pro Ser Ser Ile Lys Thr Phe Glu Glu Tyr Lys Lys Ala
      20             25             30
Phe Asn Lys Ser Tyr Ala Thr Phe Glu Asp Glu Glu Ala Ala Arg Lys
      35             40             45
Asn Phe Leu Glu Ser Val Lys Tyr Val Gln Ser Asn Gly Gly Ala Ile
      50             55             60
Asn His Leu Ser Asp Leu Ser Leu Asp Glu Phe Lys Asn Arg Phe Leu
      65             70             75             80
Met Ser Ala Glu Ala Phe Glu His Leu Lys Thr Gln Phe Asp Leu Asn
      85             90             95
Ala Glu Thr Asn Ala Cys Ser Ile Asn Gly Asn Ala Pro Ala Glu Ile
      100            105            110
Asp Leu Arg Gln Met Arg Thr Val Thr Pro Ile Arg Met Gln Gly Gly
      115            120            125
Cys Gly Ser Ala Trp Ala Phe Ser Gly Val Ala Ala Thr Glu Ser Ala
      130            135            140
Tyr Leu Ala Tyr Arg Asn Gln Ser Leu Asp Leu Ala Glu Gln Glu Leu
      145            150            155            160
Val Asp Cys Ala Ser Gln His Gly Cys His Gly Asp Thr Ile Pro Arg
      165            170            175
Gly Ile Glu Tyr Ile Gln His Asn Gly Val Val Gln Glu Ser Tyr Tyr
      180            185            190

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Arg	Tyr	Val	Ala	Arg	Glu	Gln	Ser	Cys	Arg	Arg	Pro	Asn	Ala	Gln	Arg
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Phe	Gly	Ile	Ser	Asn	Tyr	Cys	Gln	Ile	Tyr	Pro	Pro	Asn	Val	Asn	Lys
210				215				220							
Ile	Arg	Glu	Ala	Leu	Ala	Gln	Thr	His	Ser	Ala	Ile	Ala	Val	Ile	Ile
225				230				235				240			
Gly	Ile	Lys	Asp	Leu	Asp	Ala	Phe	Arg	His	Tyr	Asp	Gly	Arg	Thr	Ile
245				250				255							
Ile	Gln	Arg	Asp	Asn	Gly	Tyr	Gln	Pro	Asn	Tyr	His	Ala	Val	Asn	Ile
260				265				270							
Val	Gly	Tyr	Ser	Asn	Ala	Gln	Gly	Val	Asp	Tyr	Trp	Ile	Val	Arg	Asn
275				280				285							
Ser	Trp	Asp	Thr	Asn	Trp	Gly	Asp	Asn	Gly	Tyr	Gly	Tyr	Phe	Ala	Ala
290				295				300							
Asn	Ile	Asp	Leu	Met	Met	Ile	Glu	Glu	Tyr	Pro	Tyr	Val	Val	Ile	Leu
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ggaggtgccca tcaaccattt gtccgatttg tcgttggtatg aattcaaaaa ccgattttttg 240
atgagtgcag aagcttttga acacctcaaa actcaattcg atttgaacgc ctgcagtatc 300
aatggaaatg ctccagctga aatcgatttg cgacaaatgc gaactgtcac tcccattcgt 360
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tatttggctt accgtaatca atcattggat cttgctgaac aagaattagt cgattgtgct 480
tcccaacacg gttgtcatgg tgataccatt ccacgtggta ttgaatacat ccaacataat 540
ggtgtcgtcc aagaaagcta ctatcgatac gttgcacgag aacaatcatg ccgacgacca 600
aatgcacaac gtttcggtat ctcaaactat tgccaaattt acccaccaaa tgtaaacaaa 660
attcgtgaag ctttggctca aaccacagc gctattgccg tcattatttg catcaaagat 720
ttagacgcat tccgtcatta tgatggccga acaatcattc aacgcgataa tggttaccaa 780
ccaaactatc acgctgtcaa cattgttggt tacagtaacg cacaagggtg cgattattgg 840
atcgtacgaa acagttggga taccaattgg ggtgataatg gttacggtta ttttgcgtgc 900
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<210> 4
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<213> Artificial Sequence

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Tyr Ala Arg Pro Ser Ser Ile Lys Thr Phe Glu Glu Tyr Lys Lys Ala
20 25 30
Phe Asn Lys Ser Tyr Ala Thr Phe Glu Asp Glu Glu Ala Ala Arg Lys
35 40 45
Asn Phe Leu Glu Ser Val Lys Tyr Val Gln Ser Asn Gly Gly Ala Ile
50 55 60
Asn His Leu Ser Asp Leu Ser Leu Asp Glu Phe Lys Asn Arg Phe Leu
65 70 75 80
Met Ser Ala Glu Ala Phe Glu His Leu Lys Thr Gln Phe Asp Leu Asn
85 90 95
Ala Cys Ser Ile Asn Gly Asn Ala Pro Ala Glu Ile Asp Leu Arg Gln
100 105 110
Met Arg Thr Val Thr Pro Ile Arg Met Gln Gly Gly Cys Gly Ser Cys
115 120 125
Trp Ala Phe Ser Gly Val Ala Ala Thr Glu Ser Ala Tyr Leu Ala Tyr
130 135 140
Arg Asn Gln Ser Leu Asp Leu Ala Glu Gln Glu Leu Val Asp Cys Ala
145 150 155 160
Ser Gln His Gly Cys His Gly Asp Thr Ile Pro Arg Gly Ile Glu Tyr
165 170 175
Ile Gln His Asn Gly Val Val Gln Glu Ser Tyr Tyr Arg Tyr Val Ala
180 185 190
Arg Glu Gln Ser Cys Arg Arg Pro Asn Ala Gln Arg Phe Gly Ile Ser
195 200 205
Asn Tyr Cys Gln Ile Tyr Pro Pro Asn Val Asn Lys Ile Arg Glu Ala
210 215 220
Leu Ala Gln Thr His Ser Ala Ile Ala Val Ile Ile Gly Ile Lys Asp
225 230 235 240
Leu Asp Ala Phe Arg His Tyr Asp Gly Arg Thr Ile Ile Gln Arg Asp
245 250 255
Asn Gly Tyr Gln Pro Asn Tyr His Ala Val Asn Ile Val Gly Tyr Ser

260	265	270
Asn Ala Gln Gly Val Asp Tyr Trp	Ile Val Arg Asn Ser Trp Asp Thr	
275	280	285
Asn Trp Gly Asp Asn Gly Tyr Gly Tyr Phe Ala Ala Asn Ile Asp Leu		
290	295	300
Met Met Ile Glu Glu Tyr Pro Tyr Val Val Ile Leu		
305	310	315

<210> 5
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 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Mutant of DerP1

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 gaagatgaag aagctgcccg taaaaacttt ttggaatcag taaaatatgt tcaatcaaatt 180
 ggaggtgcca tcaaccattt gtccgatttg tcgttggatg aattcaaaaa ccgattttttg 240
 atgagtgcag aagcttttga acacctcaaa actcaattcg atttgaatgc tgaaactaac 300
 gcttgcagta tcaatggaaa tgctccagct gaaatcgatt tgcgacaaat gcgaactgtc 360
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 gtcgattgtg cttcccaaca cggttgtcat ggtgatacca ttccacgtgg tattgaatac 540
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 tgccgacgac caaatgcaca acgttttcggt atctcaaaact attgccaaat ttaccaccca 660
 aatgtaaaca aaattcgtga agctttggct caaaccacaca gcgctattgc cgtcattatt 720
 ggcatcaaag atttagacgc attccgtcat tatgatggcc gaacaatcat tcaacgcgat 780
 aatggttacc aaccaaacta tgctgctgtc aacattgttg gttacagtaa cgcacaaggt 840
 gtcgattatt ggatcgtacg aaacagtttg gataccaatt ggggtgataa tggttacggg 900
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 taa 963

<210> 6
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 <212> PRT
 <213> Artificial Sequence

<220>
 <223> Mutant of DerP1

<400> 6

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Tyr	Ala	Arg	Pro	Ser	Ser	Ile	Lys	Thr	Phe	Glu	Glu	Tyr	Lys	Lys	Ala
			20					25					30		
Phe	Asn	Lys	Ser	Tyr	Ala	Thr	Phe	Glu	Asp	Glu	Glu	Ala	Ala	Arg	Lys
	35						40					45			
Asn	Phe	Leu	Glu	Ser	Val	Lys	Tyr	Val	Gln	Ser	Asn	Gly	Gly	Ala	Ile
50						55					60				
Asn	His	Leu	Ser	Asp	Leu	Ser	Leu	Asp	Glu	Phe	Lys	Asn	Arg	Phe	Leu
65					70				75					80	
Met	Ser	Ala	Glu	Ala	Phe	Glu	His	Leu	Lys	Thr	Gln	Phe	Asp	Leu	Asn
			85					90						95	
Ala	Glu	Thr	Asn	Ala	Cys	Ser	Ile	Asn	Gly	Asn	Ala	Pro	Ala	Glu	Ile
			100					105					110		
Asp	Leu	Arg	Gln	Met	Arg	Thr	Val	Thr	Pro	Ile	Arg	Met	Gln	Gly	Gly
		115					120						125		
Cys	Gly	Ser	Cys	Trp	Ala	Phe	Ser	Gly	Val	Ala	Ala	Thr	Glu	Ser	Ala
130					135						140				
Tyr	Leu	Ala	Tyr	Arg	Asn	Gln	Ser	Leu	Asp	Leu	Ala	Glu	Gln	Glu	Leu
145					150					155					160
Val	Asp	Cys	Ala	Ser	Gln	His	Gly	Cys	His	Gly	Asp	Thr	Ile	Pro	Arg
			165					170						175	
Gly	Ile	Glu	Tyr	Ile	Gln	His	Asn	Gly	Val	Val	Gln	Glu	Ser	Tyr	Tyr
		180						185						190	
Arg	Tyr	Val	Ala	Arg	Glu	Gln	Ser	Cys	Arg	Arg	Pro	Asn	Ala	Gln	Arg
		195					200						205		
Phe	Gly	Ile	Ser	Asn	Tyr	Cys	Gln	Ile	Tyr	Pro	Pro	Asn	Val	Asn	Lys
210						215						220			
Ile	Arg	Glu	Ala	Leu	Ala	Gln	Thr	His	Ser	Ala	Ile	Ala	Val	Ile	Ile
225					230					235					240
Gly	Ile	Lys	Asp	Leu	Asp	Ala	Phe	Arg	His	Tyr	Asp	Gly	Arg	Thr	Ile
			245					250						255	
Ile	Gln	Arg	Asp	Asn	Gly	Tyr	Gln	Pro	Asn	Tyr	Ala	Ala	Val	Asn	Ile
		260						265					270		
Val	Gly	Tyr	Ser	Asn	Ala	Gln	Gly	Val	Asp	Tyr	Trp	Ile	Val	Arg	Asn
		275					280						285		
Ser	Trp	Asp	Thr	Asn	Trp	Gly	Asp	Asn	Gly	Tyr	Gly	Tyr	Phe	Ala	Ala
290					295							300			
Asn	Ile	Asp	Leu	Met	Met	Ile	Glu	Glu	Tyr	Pro	Tyr	Val	Val	Ile	Leu
305				310						315					320

<210> 7
<211> 339
<212> PRT
<213> Artificial Sequence

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20 25 30
Ala His Ser Ala Phe Ala Ala Asp Pro Arg Pro Ser Ser Ile Lys Thr
35 40 45
Phe Glu Glu Tyr Lys Lys Ala Phe Asn Lys Ser Tyr Ala Thr Phe Glu
50 55 60
Asp Glu Glu Ala Ala Arg Lys Asn Phe Leu Glu Ser Val Lys Tyr Val
65 70 75 80
Gln Ser Asn Gly Gly Ala Ile Asn His Leu Ser Asp Leu Ser Leu Asp
85 90 95
Glu Phe Lys Asn Arg Phe Leu Met Ser Ala Glu Ala Phe Glu His Leu
100 105 110
Lys Thr Gln Phe Asp Leu Asn Ala Cys Ser Ile Asn Gly Asn Ala Pro
115 120 125
Ala Glu Ile Asp Leu Arg Gln Met Arg Thr Val Thr Pro Ile Arg Met
130 135 140
Gln Gly Gly Cys Gly Ser Cys Trp Ala Phe Ser Gly Val Ala Ala Thr
145 150 155 160
Glu Ser Ala Tyr Leu Ala Tyr Arg Asn Gln Ser Leu Asp Leu Ala Glu
165 170 175
Gln Glu Leu Val Asp Cys Ala Ser Gln His Gly Cys His Gly Asp Thr
180 185 190
Ile Pro Arg Gly Ile Glu Tyr Ile Gln His Asn Gly Val Val Gln Glu
195 200 205
Ser Tyr Tyr Arg Tyr Val Ala Arg Glu Gln Ser Cys Arg Arg Pro Asn
210 215 220
Ala Gln Arg Phe Gly Ile Ser Asn Tyr Cys Gln Ile Tyr Pro Pro Asn
225 230 235 240
Ala Asn Lys Ile Arg Glu Ala Leu Ala Gln Thr His Ser Ala Ile Ala
245 250 255
Val Ile Ile Gly Ile Lys Asp Leu Asp Ala Phe Arg His Tyr Asp Gly

260	265	270
Arg Thr Ile Ile Gln Arg Asp Asn Gly Tyr Gln Pro Asn Tyr His Ala		
275	280	285
Val Asn Ile Val Gly Tyr Ser Asn Ala Gln Gly Val Asp Tyr Trp Ile		
290	295	300
Val Arg Asn Ser Trp Asp Thr Asn Trp Gly Asp Asn Gly Tyr Gly Tyr		
305	310	315
Phe Ala Ala Asn Ile Asp Leu Met Met Ile Glu Glu Tyr Pro Tyr Val		320
325	330	335
Val Ile Leu		

<210> 8

<211> 343

<212> PRT

<213> Artificial Sequence

<220>

<223> Mutant of DerP1

<400> 8

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20	25	30
Ala His Ser Ala Phe Ala Ala Asp Pro Arg Pro Ser Ser Ile Lys Thr		
35	40	45
Phe Glu Glu Tyr Lys Lys Ala Phe Asn Lys Ser Tyr Ala Thr Phe Glu		
50	55	60
Asp Glu Glu Ala Ala Arg Lys Asn Phe Leu Glu Ser Val Lys Tyr Val		
65	70	75
Gln Ser Asn Gly Gly Ala Ile Asn His Leu Ser Asp Leu Ser Leu Asp		80
85	90	95
Glu Phe Lys Asn Arg Phe Leu Met Ser Ala Glu Ala Phe Glu His Leu		
100	105	110
Lys Thr Gln Phe Asp Leu Asn Ala Glu Thr Asn Ala Cys Ser Ile Asn		
115	120	125
Gly Asn Ala Pro Ala Glu Ile Asp Leu Arg Gln Met Arg Thr Val Thr		
130	135	140
Pro Ile Arg Met Gln Gly Gly Cys Gly Ser Ala Trp Ala Phe Ser Gly		
145	150	155
Val Ala Ala Thr Glu Ser Ala Tyr Leu Ala Tyr Arg Asn Gln Ser Leu		160

	165		170		175										
Asp	Leu	Ala	Glu	Gln	Glu	Leu	Val	Asp	Cys	Ala	Ser	Gln	His	Gly	Cys
	180		185		190										
His	Gly	Asp	Thr	Ile	Pro	Arg	Gly	Ile	Glu	Tyr	Ile	Gln	His	Asn	Gly
	195		200		205										
Val	Val	Gln	Glu	Ser	Tyr	Tyr	Arg	Tyr	Val	Ala	Arg	Glu	Gln	Ser	Cys
210			215		220										
Arg	Arg	Pro	Asn	Ala	Gln	Arg	Phe	Gly	Ile	Ser	Asn	Tyr	Cys	Gln	Ile
225			230		235									240	
Tyr	Pro	Pro	Asn	Ala	Asn	Lys	Ile	Arg	Glu	Ala	Leu	Ala	Gln	Thr	His
			245		250									255	
Ser	Ala	Ile	Ala	Val	Ile	Ile	Gly	Ile	Lys	Asp	Leu	Asp	Ala	Phe	Arg
	260		265		270										
His	Tyr	Asp	Gly	Arg	Thr	Ile	Ile	Gln	Arg	Asp	Asn	Gly	Tyr	Gln	Pro
	275		280		285										
Asn	Tyr	His	Ala	Val	Asn	Ile	Val	Gly	Tyr	Ser	Asn	Ala	Gln	Gly	Val
	290		295		300										
Asp	Tyr	Trp	Ile	Val	Arg	Asn	Ser	Trp	Asp	Thr	Asn	Trp	Gly	Asp	Asn
305			310		315									320	
Gly	Tyr	Gly	Tyr	Phe	Ala	Ala	Asn	Ile	Asp	Leu	Met	Met	Ile	Glu	Glu
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Tyr	Pro	Tyr	Val	Val	Ile	Leu									
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<210> 9
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<220>
 <223> Primer

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36

<210> 10
 <211> 28
 <212> DNA
 <213> Artificial Sequence

<220>
 <223> Primer

<400> 10
ggcgttagta gcttcagcct ctcttttc

28

<210> 11

<211> 86

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 11

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attagctgct ccaactaacg cctgca 86

<210> 12

<211> 78

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 12

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aggaaatctc atcgtttg 78

<210> 13

<211> 74

<212> DNA

<213> Artificial Sequence

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<223> Primer

<400> 13

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tttgaacgcc tgca 74

<210> 14

<211> 66

<212> DNA

<213> Artificial Sequence

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<210> 15

<211> 25

<212> DNA

<213> Artificial Sequence

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<223> Primer

<400> 15

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<210> 16

<211> 24

<212> DNA

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<211> 113

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 17

taatggaaat gtcacagctg aaatcgattt gcgacaaatg cgaactgtca ctcccattcg 60
tatgcaagga ggctgtgggt cagcttgggc tttctctggt gttgccgcaa ctg 113

<210> 18
<211> 114
<212> DNA
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<223> Primer

<400> 18
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<210> 19
<211> 75
<212> DNA
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atttgaacgc ctgca 75

<210> 20
<211> 67
<212> DNA
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<220>

<223> Primer

<400> 20
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gtttttg 67

<210> 21
<211> 25
<212> DNA
<213> Artificial Sequence

<220>

<223> Primer

<400> 21

catgaaaatt gttttggcca tcgcc

25

<210> 22

<211> 24

<212> DNA

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<223> Primer

<400> 22

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24

<210> 23

<211> 78

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

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78

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<211> 70

<212> DNA

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<220>

<223> Primer

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<210> 25

<211> 172

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 25

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tatgctacct tegaagatga agaagctgcc cgtaaaaact ttttggaatc agtaaaatat 120
gttcaatcaa atggaggtgc catcaaccat ttgtccgatt tgcgttgga tg 172

<210> 26

<211> 172

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 26

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tttactgatt ccaaaaagtt tttacgggca gcttcttcat cttcgaaggt agcataactt 120
ttgttgaagg ctttttttga ttcttcaaaa gttttgatcg atgacggccg gg 172

<210> 27

<211> 17

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

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<213> Artificial Sequence

<220>

<223> Primer

<400> 29

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atttgaacgc ctgca 75

<210> 30

<211> 67

<212> DNA

<213> Artificial Sequence

<220>

<223> Primer

<400> 30

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gttttttg 67